Nematodes of the Subfamily Splendidofilariinae in the Subcutis and Coelomic Cavity of a Large Day Gecko, *Phelsuma madagascariensis grandis*

K. Michelle Haag[1,3], DVM, Cheryl B. Greenacre[1,4], DVM, DABVP-Avian, Ryan S. Devoe[1,5], DVM, DACZM, Dana Ambrose[2], MS

1. Department of Small Animal Medicine, College of Veterinary Medicine, University of Georgia, Athens, GA 30602, USA

2. Department of Parasitology, College of Veterinary Medicine, University of Georgia, Athens, GA 30602, USA

3. Current address: Commerce Veterinary Hospital, 1012 S. Broad St., Commerce, GA 30529, USA

4. Department of Small Animal Clinical Sciences, College of Veterinary Medicine, The University of Tennessee, Knoxville, TN 37996, USA

5. Dallas Zoo, 650 South R.L. Thornton Freeway, Dallas, TX 75203, USA

ABSTRACT: A presumably captive-born, 3-year old, female large day gecko, *Phelsuma madagascariensis grandis*, presented with a subcutaneous mass that contained a nematode of the subfamily Spendidofilariinae. Eleven months later the gecko presented for anorexia and depression 28 d post egg-laying. Radiographs showing two poorly calcified eggs were suggestive of dystocia and an exploratory laparotomy was performed. Severe, non-septic coelomitis associated with nine nematodes in granulomatous capsules and a retained egg were discovered. All nematodes were found to be in the subfamily Splendidofilariinae that have not been reported previously in day geckos.

KEY WORDS: day gecko, *Phelsuma madagascariensis grandis,* Splendidofilariinae, nematode, coelomitis, subcutaneous.

INTRODUCTION

The native habitat for large day geckos, *Phelsuma madagascariensis grandis*, are islands in the Indian Ocean, especially Madagascar. Although an increasing number of various species of day geckos are bred in captivity, most are wild caught and imported. There are about 60 species and subspecies of day geckos.

The subfamily Spendidofilariinae is comprised of two genera of nematodes, *Thamugadia* and *Madathamugadia*. Like other filarids, they produce microfilaria and require bloodsucking arthropods as intermediate hosts and vectors.

Madathamugadia spp. are parasites of gerrhosaurids and iguanids of Madagascar, Turkmenistan, and Africa, comprising eight species: *M. zonosauri, M. hoplurii, M. ivaschkini, M. wanjii, M. versterae, M. bissani, M. ineichi*, and *M. huambensis*. *Madathamugadia ivaschkini* was previously classified in the genus *Thamugadia* and has been identified as a parasite of geckonids. *Madathamugadia* spp. are characterized by a divided esophagus. Males have a short tail, a double row of genital papillae cranial to the cloaca, and asymmetrical spicules. Females have a vulva cranial to the esophagus. Adult nematodes have been found in the coelomic cavities of their hosts. The microfilaria are small, approximately 50 μm long, and present in the blood of the host (Anderson, 1992, Bain, 1993, Sood, 1999).

Thamugadia spp. are found in geckonids of North Africa and India, agamids in Lebanon, and lacertids and agamids in Turkmenistan. There are five species in the genus, *T. hyalina, T. agamae, T. wertheimae, T. hemidacyla, and T. skrjabini. Thamugadia* spp. have a smooth cuticle and narrow and indistinct lateral fields. Lateral fields are an interruption of the transverse striae by longitudinal cuticular thickenings located above the lateral chords. The mouth is simple, without lips, but having six small papillae. The esophagus is short and may be variably divided. The tail of both sexes is long and digitiform. In males, caudal alae

and genital papillae are absent in one species, *T. hyalina*. In two of the species, *T. agamae* and *T. skrjabini*, there is a double row of caudal alae posterior to the cloaca. The females are opisthodelphys, meaning the uterus (or uteri) is directed posteriorly, and the vulva is located posterior or anterior to the esophagus. Adults are found in the subcutis of their hosts. The microfilaria are 122-140 µm in length, sheathed, and present in the blood of the host (Anderson, 1992, Bain *et al*, 1992, Bain *et al*, 1993, Sood, 1999).

Adult *Madathamugadia* and *Thamugadia* filarids can only be differentiated on the basis of characteristics of the male worms. Species of the genera are differentiated by microfilaria characteristics.

Case Report

A 3-year old, 46 g, presumably captive-born large day gecko was purchased at a reptile fair and it was initially housed alone. Fifteen months after purchase, the gecko presented for a dorsolateral, subcutaneous vermiform mass cranial to the left rear leg (Figure 1). The skin over the mass was sterilely prepped with chlorhexadine scrub (Nolvasan Surgical Scrub, Fort Dodge Animal Health, Fort Dodge, IA 50501) and 70% alcohol. Prior to incision, ice was placed over the mass to numb the area, after which alcohol was applied again. While the gecko was manually restrained, two mm of skin over the mass was lanced with a #15 blade. The gecko did not appear to be in pain either during or after creating the incision. An off-white nematode, 0.8 cm long, was removed with forceps (Figure 2 and 3). The area was flushed with 5 ml of sterile saline, dried, and 1 drop of cyanoacralate tissue glue (Nexaband Liquid, Veterinary Products Laboratory, Phoenix, AZ) applied for skin closure. Approximately 0.02 ml of blood was taken from the ventral coccygeal vein. Blood smears were made, but no microfilaria were seen. No anthelmintics were administered.

Five months after removal of the subcutaneous filarid, a nematode was found in the gecko's feces, that on evaluation, was found to be an ascarid. The gecko was treated with 50mg/kg oral fenbendazole (Panacur, DPT Laboratories, San Antonio, TX). The treatment was repeated 14 d later. One month after the two doses of fenbendazole were given, the female was placed in an enclosure with a male large day gecko.

During the next five months, the gecko had four clutches of eggs (two eggs per clutch), with approximately one month between clutches. Twenty-eight days after the fourth clutch, the gecko was found lying on the bottom of the enclosure. The gecko was thought to be anorexic and depressed post egg-laying. The gecko was lethargic, anorexic, had an enlarged coelomic cavity and weighed 43 g. No eggs could be palpated on presentation. Radiographs indicated two poorly calcified areas in the caudal coelomic cavity. An attempt was made to stimulate the gecko to lay the presumed eggs by removal from the male, increasing the cage temperature from 30°C (85°F) to 32°C (90°F) and the humidity from 75% to 90%. Oral calcium glubionate (Calciquid, Econolab, Westland, MI) was administered at 0.01ml (10 ml/kg) every 12 hr for 5 d.

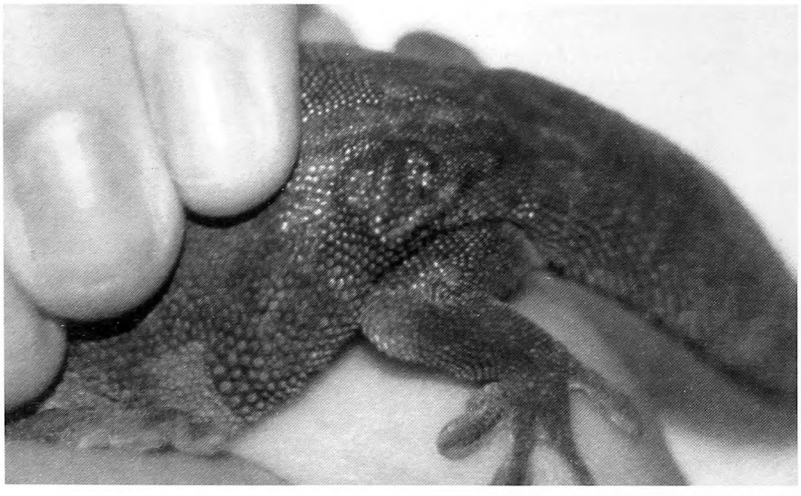

Figure 1- A 3-year old large day gecko, *Phelsuma madagascariensis grandis,* with a subcutaneous vermiform mass on the lateral body wall 0.5 cm cranial to the left rear leg.

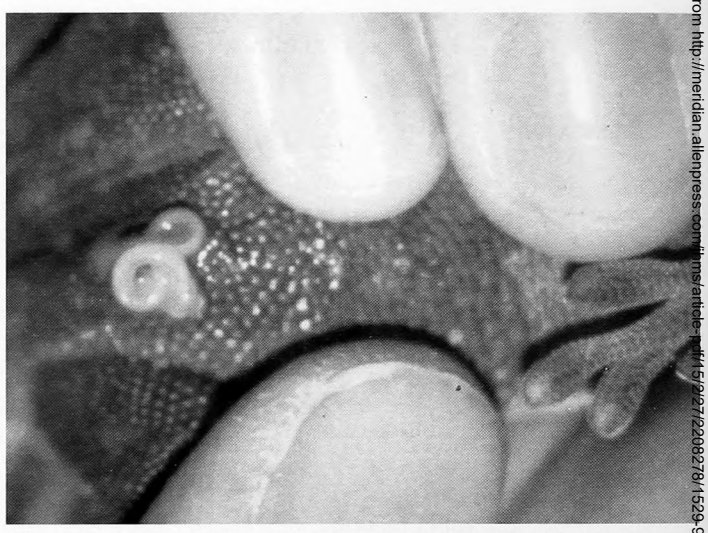

Figure 2. Removal of a subcutaneous nematode, *Madathamugadia* or *Thamugadia* spp., from a large day gecko, *Phelsuma madagascariensis grandis*, after a 0.2 cm skin incision was made.

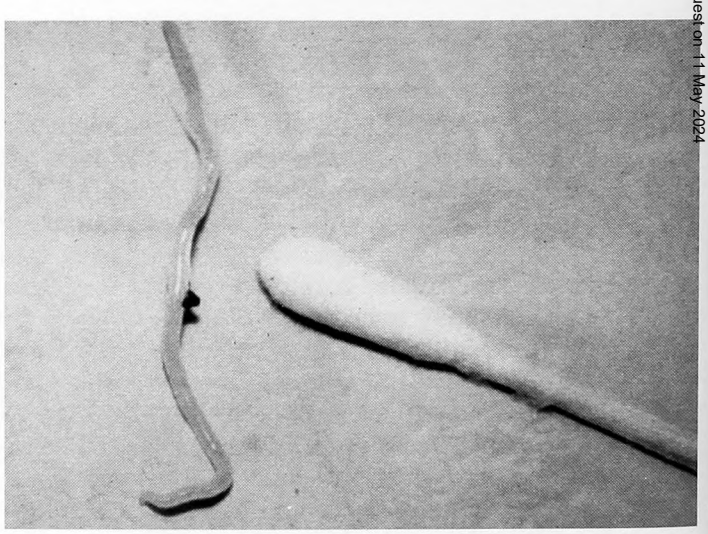

Figure 3 – The subcutaneous nematode, *Madathamugadia* or *Thamagadia* spp., removed from a subcutaneous mass on a day gecko, *Phelsuma madagascariensis grandis*, was 0.8 cm in length and off-white to slightly yellow in color.

After five days, the gecko (46 g) had not passed any eggs and it had grown more lethargic. The gecko was prepared for an ovariosalpingectomy and received 2 mg/kg butorphanol (Torbugesic, Fort Dodge Animal Health, Fort Dodge, IA) SC for pain management. It was induced with isoflurane (Aerrane, Anaquest, Inc., Liberty Corner, NJ) delivered through a vaporizor. The gecko was maintained on isoflurane by assisted ventilation one to two times per minute via an endotracheal tube. A water recirculating heating pad was placed under the gecko during anesthesia to maintain body temperature. A 2 cm right paramedian incision was made and several granulomatous capsules containing filarid worms were discovered in the coelomic cavity. The capsules were individually lanced and a total of nine filarids were recovered and placed in 70% alcohol. Some filarids were necrotic and dead. One egg was found in the right oviduct. The left body wall was adhered multifocally to underlying organs, including the left surface of the liver lobe. The surface of the liver was irregular. Biopsies of organs were not taken for histopathology. The gecko was given 33 ml/kg ml warmed 2.5 % dextrose with half-strength Lactated Ringers Solution (LRS, USP, B. Braun Medical Inc, Irvine, CA) SC. Bipolar electrocautery, 5-0 PDS (Ethicon, Cornelia, GA), and a small ligaclip were used for ligation of small vessels incised during the procedure. The coelomic muscle was sutured with 5-0 PDS (Ethicon, Cornelia, GA) in a simple continuous pattern and the skin in an everting pattern.

The gecko was sent home after it had recovered from surgery. The calcium glubionate was continued for 14 d post-surgery. The gecko was also given 5 mg/kg enrofloxacin (Baytril injection, Bayer Corporation, Agricultural Division, Animal Health, Shawnee Mission, KS) orally every 12 hr for 14 d and lactulose at 2 mg/kg every 24 hr for 14 d. Peach and apricot baby food and water were fed to the gecko by syringe. The gecko was kept in a plastic container with paper towels for substrate. The temperature was maintained at 30°C (85°F) to 32°C (90°F) and humidity at 65 – 75%, measured with a Precision Analog Humidity Gauge (Zoo Med Laboratories Inc, San Luis Obispo, CA), by misting the enclosure several times a day. Although the surgical incision healed well, the gecko had continued anorexia and weight loss. Every two to three days for two weeks, 109 ml/kg of 0.9% sodium chloride (Saline solution 0.9%, B. Braun Medical Inc, Irvine, CA) was administered SC with no improvement.

The presence of further nematodes was thought to be contributing to the gecko's lack of improvement, therefore three weeks post-surgery, the gecko was administered a dose of oral ivermectin (Ivomec 1% injection for cattle and swine, Merck AgVet Division, Merck and Co. Inc; Rahway, NJ) at 0.2 mg/kg. The following day the gecko was found dead. On necropsy, 25 filarid worms were discovered in granulomatous capsules in the left cranial coelomic cavity adhered to the left liver lobe, where the area of adhesions was observed at surgery. The worms were dead and poorly preserved. A non-septic coelomitis was noted and the liver surface appeared irregular.

The male gecko, housed with this female, continues to thrive and has shown no clinical abnormalities. Direct fecal smears and zinc sulfate fecal floatations from this animal on three different examinations spanning three years have been negative for protozoan cysts or nematode larvae. Blood smears were performed on two of these visits and were found to be negative for microfilaria.

Discussion

Many of the worms found at necropsy were poorly preserved and could not be used for identification. All well-preserved worms were females. Without a male worm, it was not possible to determine whether the gecko was infected with species of *Madathamugadia*, *Thamugadia,* or both. Neither of these genera have been described as parasites of the large day gecko. *Madathamugadia* spp. are found in the same locality, Madagascar, as large day geckos, but have only been described in plated lizards (subfamily Gerrhosaurinae, family Cordylidae) and iguanids (family Iguanidae) in that region. *Thamugadia* spp. have been found in many species of geckos, but not in Madagascar. If microfilaria had been obtained, the genus and/or species of worm might have been determined. It is important to place nematodes in alcohol, as was done in this case, rather than formalin so that identifiable structures are preserved, enabling taxonomic identification.

The gecko in this case was presumably captive-born, though attempts to locate the breeders were unsuccessful, so it is not possible to know whether this gecko was actually wild-caught. History of contact with other lizard species prior to purchase is unknown. Location of the gecko at the time of infection cannot be determined. Wild reptiles often have heavy parasite burdens without showing clinical disease. In captivity, parasites cause increased incidence of disease due to increased stress (Frye,1991). This gecko showed no signs of disease until development of a subcutaneous mass. After the subcutaneous nematode was removed, the gecko successfully laid 4 clutches of eggs. The parasite burden and the stress of egg laying were felt to be significant in the cause of death of the gecko.

The gecko may have died of an anaphylactic reaction to the parasites in its coelomic cavity after treatment with ivermectin. Most of the nematodes found at necropsy were in granulomatous capsules on the left coelom dorsal to the liver. Adult filarids in the coelomic cavity or in the subcutis of chameleons are very resistant to treatment (Barrie, 1999); viable nematodes have been identified at necropsy despite several anthelmintic treatments. If all parasites had been removed during surgery, the prognosis of this case might have been better. The best means of preventing disease from filarid parasites is to prevent transmission of the microfilaria into the host. The presence of infection in wild-caught animals should be considered.

Management of this case involved numerous diagnostic and therapeutic methods; however, more successful results may have been achieved with alternative or additional modalities. For example, ice was chosen as the local anesthetic for removal of the first subcutaneous parasite. The gecko should have been briefly anesthetized and/or provided with an analgesic since this procedure may have been painful and pain can be difficult to assess in reptiles. Also, a more extensive exploratory laparotomy, including a liver

biopsy could have been performed, but risk of fatal hemorrhage was a concern. Ultrasound could have been a useful diagnostic tool to locate the nematodes and assess the internal organs.

This paper describes severe nematode infection in a large day gecko. The nematodes were found to be in the subfamily Splendidofilariinae which have not been previously reported in day geckos. It is unclear if the gecko acquired this infection in the United States or in Madagascar.

References

Anderson RC. 1992. The Superfamily Filarioidea. *In* Nematode Parasites of Vertebrates: Their Development and Transmission. University Press, Cambridge, UK:437- 493.

Bain O, Petit G, Paperna I, Finkelman S, Killick-Kendrick M.1992. A new filaria of a lizard transmitted by sandflies. Mem. Inst. Oswaldo Cruz, Rio de Janeiro:87 (1):21-29.

Bain O, Wanji S, Petit G, Paperna I, Finkelman S.1993. Filaires Spendidofilariinae de lézards: nouvelles espèces, redescription, cycle chez phlébotome. Systematic Parasitology, 26(2):97-15

Barrie MT. 1999. Chameleon Medicine. *In* Fowler ME and Miller RE (eds): Zoo and Wild Animal Medicine, Current Therapy. W B Saunders Co., Philadelphia, PA:200-204.

Frye FL. 1991. Applied Clinical Nonhemic Parasitology of Reptiles *In* Biomedical and Surgical Aspects of Captive Reptile Husbandry, 2nd edition, Vol. 1, Krieger Publishing Co. Malabar, FL:296-314.

Sood, ML. 1999. Superfamily Filarioidea. *In* Reptile Nematodes from South Asia. Milton Book Co. Dehradun, India:173-179.